Abstract  Large-bodied zooplankton of the genus *Daphnia* are thought to be keystone species in freshwater pelagic habitats, potentially able to exert strong grazing effects and enhance phosphorus limitation of algae. I examined the degree to which *Daphnia pulex* differ from small-bodied zooplankton in their effects on algal biomass, seston C:P and N:P, total nitrogen and total phosphorus. This was done with both survey data from natural ponds and an in situ experiment in which *D. pulex* was compared to a small zooplankton assemblage under low and high nutrient conditions and in two different ponds. *D. pulex* effects on algae were only evident under high nutrient conditions. In natural ponds, *D. pulex* dominance resulted in a significantly weaker chlorophyll–total phosphorus relationship, with the divergence between *D. pulex* and small zooplankton-dominated systems being greatest in highly enriched ponds. In the experiment, *D. pulex* exerted stronger top-down control in enriched treatments only and tended to graze algae to lower levels in the more productive pond. Dynamics of C:P over the course of the experiment did not reveal strong effects of zooplankton composition. However, data on the final date of the experiment provided some evidence that *D. pulex* can enhance phosphorus limitation of algae; total phosphorus was lower and C:P higher in *D. pulex* treatments. Survey results revealed no effects of *D. pulex* on seston C:P or N:P, suggesting that this species may not be an important factor governing phosphorus limitation of algae in natural ponds.

Keywords  Algae · Keystone species · Nutrient cycling · Stoichiometry · Zooplankton

Introduction

Recently the field of ecology has seen an upsurge of interest in the effects of species, species richness and functional groups on ecosystem function and stability (Schulze and Mooney 1993; Naeem et al. 1994; Tilman and Downing 1994; McGrady-Steed et al. 1997; Hulot et al. 2000). This research has been spurred in large part by practical needs, for gaining insight into such relationships undoubtedly holds important implications for how we manage natural resources as well as our ability to predict the outcome of environmental impacts and species losses. Traditionally, important or "keystone" species have been distinguished by their inordinately large effects on community properties relative to other species. Such key species may exact effects through their ability to dominate community biomass (a numerical effect) or through extraordinarily high per capita effects. While conventionally measured at the level of communities (e.g., effects on species composition or richness), the presence of such species could have important repercussions on ecosystem-level properties, causing the relationship between species richness and ecosystem function to be disjunct (Power et al. 1996). It is evident that our ability to comprehend and predict the consequences of variable species composition on ecosystem properties may depend vitally on identification and experimental validation of potential keystone species (Power et al. 1996).

In temperate freshwater systems, zooplankton body size is often considered to be an important determinant of planktonic community and ecosystem properties (e.g., Pace 1984; Vanni 1987; Elser et al. 1988; Mazumder 1994a, b; Schindler et al. 1997; Cottingham 1999; Tessier et al. 2001). Commonly, large-bodied zooplankton of the genus *Daphnia* are thought to control total primary production more effectively than other zooplankton taxa, such as small-bodied cladocera or copepods (Leibold 1989; Mazumder 1994a; Cottingham and Schindler 2000), earmarking *Daphnia* as key components of trophic cascades, successful bio-control of algal blooms and system stability (by buffering nutrient per-
turbations). Recently, attention has also focused on the carbon, phosphorus and nitrogen composition (C:N:P) of zooplankton and the implications of among species variation in elemental stoichiometry on nutrient recycling. With few exceptions, Daphnia tissues appear to have the lowest C:P and N:P ratios among freshwater zooplankton (Andersen and Hessen 1991; Sterner et al. 1992). Consequently, Daphnia may sequester greater amounts of phosphorus relative to nitrogen, leading to higher nitrogen to phosphorus excretion rates compared to less phosphorus demanding taxa (Sterner et al. 1992; Elser and Urabe 1999).

Taken together, these attributes single out Daphnia as a potential keystone species whose presence can have significant impacts on the larger aquatic community and its ecosystem-level properties. While suspected, the reality of this assertion has seldom been tested via direct, long-term experimental manipulation of Daphnia in the field; rarer still are studies that compare monocultures of Daphnia with alternate taxa. A number of lake studies have compared nitrogen and phosphorus excretion by Daphnia versus zooplankton species with higher N:P ratios (e.g., copepods). These studies have largely confirmed theorized predictions; Daphnia recycle nitrogen at greater rates than phosphorus (reviewed in Elser and Urabe 1999). However, experimental elucidation of the biological relevance at the ecosystem level of such differential recycling (i.e., actual effects on algal assemblages in regards to elemental composition or nutrient limitation) is still in its formative stages (Elser and Urabe 1999). Furthermore, there have been remarkably few long-term investigations that have directly manipulated zooplankton composition with the intent of examining differential effects on algal production. Frequently, studies that have “manipulated” Daphnia presence or zooplankton size-structure have done so indirectly by manipulating the presence of planktivorous fish (e.g., Mazumder 1994b; Cottingham and Schindler 2000 and the studies analyzed in Sarnelle 1992a and Cottingham 1999), thus introducing potential confounding effects and experimental artifacts (e.g., nutrient recycling by fish; Vanni and Layne 1997). Those investigations that have directly manipulated zooplankton composition have found highly variable effects of Daphnia presence on algal standing crop, with some experiments finding small or insignificant differences (Turner and Mittelbach 1992; Brett et al. 1994), some revealing strong effects (Schoenberg and Carlson 1984; Sarnelle 1993; Tessier et al. 2001), and some finding effects that were highly variable or dependent on ecological setting (Vanni 1984; Leibold and Wilbur 1992; Steiner 2001a; reviewed in Leibold et al. 1997). The specific circumstances that mediate the strength of Daphnia grazing effects are poorly known. Yet, a number of studies, both experimental and observational, suggest that Daphnia may exert stronger effects under nutrient-enriched conditions (Vanni 1984; Sarnelle 1992a; Mazumder 1994a; Steiner 2001a). Moreover, our understanding of Daphnia impacts has been largely limited to studies of lakes. The functional role that these species play in shallow ponds is generally unknown.

In the following paper I report on an experiment in which I assessed the effects of Daphnia pulex on pond ecosystem function – specifically algal standing crop, nitrogen and phosphorus content at the scale of the entire water column (total nitrogen, TN, and total phosphorus, TP), and seston C:P ratios as indicators of phosphorus supply rates and phosphorus limitation of algal growth (Healey and Hendzel 1980). D. pulex alone was compared to a diverse assemblage of taxa composed of small-bodied cladocera, copepods, and rotifers. The experiment was performed in two fishless ponds that varied naturally in production, and under experimentally enriched and unenriched conditions. To further investigate the effects of zooplankton composition and Daphnia dominance, I also surveyed natural ponds to examine the natural relationships between D. pulex abundance and algal standing crop and seston C:N:P.

### Materials and methods

#### Field experiment

The experiment was performed in August of 1998, in two fishless ponds (P12 and P14) at the Kellogg Biological Station (hereafter K.B.S.), experimental pond facility (Hickory Corners, Mich.). Both ponds are 1.6 m deep at their centers, with a surface area of approximately 700 m². Though both ponds were fishless, zooplankton composition in the two differed greatly in the year of the study. In P12, D. pulex was present and dominant for the majority of the summer, whereas the P14 zooplankton community exhibited a seasonal loss of D. pulex and was dominated by calanoid copepods and small-bodied cladocera (primarily Diaphanosoma brachyurum) during the entire growing season. P12 was also the more productive of the two, harboring higher chlorophyll a (6.52 vs 2.62 µg l⁻¹), zooplankton biomass (0.342 vs 0.048 mg l⁻¹, dry weight) and TP (25.7 vs 16.3 µg l⁻¹) at the time of the experiment.

The experiment was performed using 1,200-l polyethylene “bag” enclosures, sealed at their bottoms to exclude sediments, and suspended in the water column using floating frames. Bags were 1 m in diameter, extended to pond bottoms, and were screened with gray fiberglass window screening on their tops to exclude insects. Though screening may have reduced sun light in the bags (potentially reducing seston C:P), effects were assumed equal among all enclosures. To explore effects of zooplankton composition, two treatments were employed: Daphnia pulex alone and an assemblage of copepods, rotifers, and small-bodied cladocera alone (hereafter collectively referred to as “small” or “small-bodied” zooplankton for simplicity). D. pulex was chosen since it is the dominant Daphnia species found in permanent, fishless ponds in southwest Michigan (Steiner 2001b). The design was executed in both P12 and P14 concurrently. To explore the interaction of enrichment and zooplankton composition on algal biomass, “D. pulex” and “small zooplankton” treatments were further crossed with two nutrient treatments (“low” and “high”) in P14 only. Manipulations consisted of additions of phosphorus (as Na₂HPO₄) with high nutrient treatments receiving an initial concentration of 75 µg P l⁻¹ and low nutrient treatments receiving no additions for an initial concentration of 16 µg P l⁻¹. Nitrogen (as NaNO₃) was added with phosphorus in a 70:1 N to P molar ratio, matched to that of the ambient pond water (based on TN:TP measurements made earlier in the growing season). Nutrients were added as a single pulse at the start of the experiment. All treatments were replicated three times for a total of 18 enclosures. All bags were filled by pumping water from their respective ponds, at